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ENDOCRINE GLANDS

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DETECTING INTERNAL SECRETION, WITH EVIDENCE FOR ACCESSORY
ACCELERATOR FIBERS FROM THE THORACIC SYMPATHETIC CHAIN

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From the Laboratories of Physiology in the Harvard Medical School

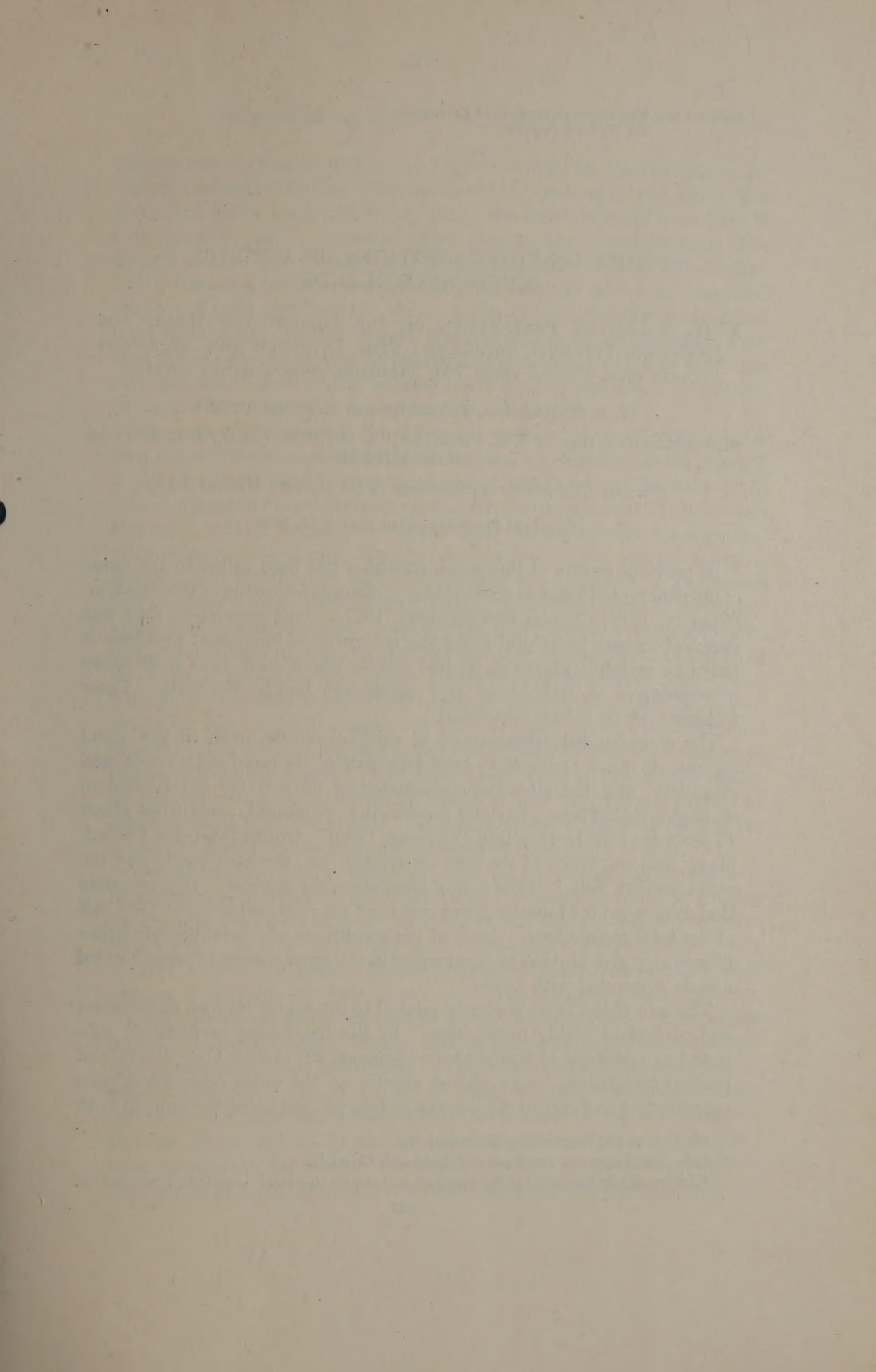
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XVII. A LASTING PREPARATION OF THE DENERVATED HEART FOR DETECTING INTERNAL SECRETION, WITH EVIDENCE FOR ACCESSORY ACCELERATOR FIBERS FROM THE THORACIC SYMPATHETIC CHAIN

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In previous papers of this series attention has been called to the value of the denervated heart as an indicator of changes occurring in the organism (Cannon, 1919; Cannon and Rapport, 1921). Our conviction that this preparation may be helpful in solving a number of important problems is based on certain features of cardiac muscle which mark it as a structure representative of widespread and significant bodily elements. These features may be briefly mentioned.

The cross-banded arrangement of cardiac muscle, even in the nodal regions, its quick response to brief stimulation, its rapid contraction and relaxation, and the all-or-none character of its activity are typical of skeletal muscle fibers. Skeletal muscle, it is estimated, constitutes about 43 per cent of the body weight (Vierordt, 1906). Unlike fat, bone, tendon, blood, skin and much of the nervous system (i.e., the fiber tracts and the nerve trunks), muscle is the seat of large chemical changes. It is probable that even when the muscles are at rest they are responsible for at least half of the total metabolism. Some of the conditions which influence cardiac muscle may give hints as to occurrences in the great masses of cross-banded muscle connected with bones.

The rate of the heart is closely related to the rate of its own metabolism and of general bodily metabolism. In the heart-lung preparation subjected to variations of temperature—between 32° and 39°C.—“the rate of gaseous metabolism varies almost exactly as the pulse rate, the oxygen consumption and carbon dioxide production *per beat* being the same at both

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temperatures" (Evans, 1912). As has been shown by observations made during prolonged fasting (Benedict, 1915), prolonged restricted diet (Benedict, Miles, Roth and Smith, 1919), the standard basal state (Harris and Benedict, 1919), muscular effort (Smith, 1922), and diseases with altered metabolism (Sturgis and Tompkins, 1920; Minot and Means, 1924), there is a fairly close relationship between the rate of the heart and the rate of total metabolism. Since cardiac rate, cardiac metabolism and total metabolism are thus bound together in the intact organism, even though the nervous system be intermediary, such influences as affect the rate and metabolism of the isolated heart may be significant in relation to metabolic changes in other muscular structures.

The rate of the heart isolated from the central nervous system is influenced only by changes of temperature, and by chemical agents brought to it in the blood stream—secretions of ductless glands and possibly products of digestion or metabolism—acting on the pace maker (Cannon and Rapport, 1921). Rogoff has argued that the denervated heart cannot yield reliable information because stimulation of the adrenal medulla, the liver and the thyroid, and also injection of extracts of various tissues will all cause a faster beat (Rogoff, 1924). One might quite as justifiably argue that intestinal muscle, used by him to determine adrenal secretion, is unreliable because heating the medium that surrounds it, or applying an excess of carbon dioxide, causes it to relax, just as adrenin does. With both indicators what is permitted to act on them is the important matter. When we have excluded action of the liver and thyroid and have not injected extracts, and then have noted that reflex acceleration, for example, occurs, but that it does not occur after inactivation of chromophil organs, an inference that the acceleration might be due to the liver, thyroid or organ extracts would be absurd. The only inference warranted is that the acceleration was due to increased activity of chromophil organs.

The denervated heart, therefore, may be reasonably considered as a representative portion of muscular tissue, probably capable of yielding insight into responses of both the striated and the non-striated forms of that tissue. It is a continually and rhythmically active muscle, living isolated in the body fluids. Its rate is fixed by influences affecting a small portion of its structure, the sinus node. The changes in rate, however, are capable of revealing alterations in the bathing fluids, (the "*milieu interne*," to use Bernard's phrase), and, as accumulating evidence tends to prove, revealing such alterations as influence contractile structures in general and also in specific ways. In short, it acts like an isolated organ perfused with different fluids—only it undergoes an *internal perfusion*. Its rate of action can be readily recorded electrically or by placing a tambour against the chest. A valuable addition to our methods, therefore, would be a preparation of the denervated heart under surgical condi-

tions so that animals thereafter could be kept alive for a considerable period and in normal health, for study of conditions influencing the rate of beat.

It appears that the only investigator who has hitherto reported having made such a preparation for the study of cardiac function is Friedenthal (1902). He destroyed the middle and lower roots of the left vago-accessory nerves at the medulla, and severed the right vagus below the recurrent branch. Then he removed the lower cervical and upper thoracic ganglia of the sympathetic chain. One dog and several rabbits survived the operation and were observed. The main features emphasized in the report on the dog were that the resting pulse was little changed from the normal, that the cardiac nerves are not essential for existence (the animal lived 8 months after the isolation was completed), and that capacity for muscular work (i.e., running) was much reduced. Unfortunately Friedenthal did not record the pulse. His statement that the rate of the denervated heart can still be markedly influenced by changes of blood pressure (which is incorrect (see Cannon and Rapport, 1921)) might indicate that the rate was not steady. As will be shown later, the conditions governing the heart are much more complicated than he realized, and the only proof of isolation from the central nervous system is relative indifference to disturbing conditions after humoral factors have been excluded. Friedenthal should have studied carefully the heart rate under different circumstances. This he apparently did not do.

In our experiments we have made use of the cat.

The innervation of the heart in the cat. The scattered distribution of nerves near the heart renders practically impossible any satisfactory denervation of the organ by operating in that region. It is necessary to find a place where the relations are simpler. At first we thought we might open the pericardium and by cutting and applying phenol we might destroy the nerves as they course along the vessels which connect the base of the heart with the pericardial wall. Our attempts were only partially successful, however, and we therefore turned elsewhere. The other situation in which the relations are relatively simple is at the origin of the cardiac nerves—the vagus trunks and the stellate ganglia. The chief difficulty with any simple exclusion of vagal influences, as by section of the nerves in the neck, lies in the consequent serious disturbances of respiration and digestion. To avoid these troubles one recurrent laryngeal nerve must be preserved in order that the larynx may act properly and protectively, and one vagus trunk must pass to the abdomen in order that the digestive functions may continue normal.

Some preliminary experiments showed that it is possible to exclude the influence of the right vagus by cutting strands which reach out from it towards the heart near the mouth of the superior vena cava and the

azygos vein. Similar strands on the left side, however, are much more readily found and severed. Because of the complexity due to the presence of the azygos vein on the right side, therefore, we selected the more readily accessible left vagus trunk for service to the gastro-intestinal canal. Investigation proved that the left recurrent laryngeal must be cut; that made necessary the preservation of the right recurrent.

Careful tests in acute experiments and also in numerous surgical operations have shown that the *right vagus* gives off all its inhibitory cardiac branches below the origin of the recurrent laryngeal (see fig. 1). Boehm and Nussbaum (1875) describe a very fine strand leaving the right vagus above the loop of the recurrent laryngeal and, having an independent course till it joins filaments from the stellate ganglion, forms with them a "common cardiac nerve." This fine strand corresponds to the right depressor nerve, as described by Perman (1924). The right recurrent laryngeal also, as a rule, sends off a twig to the common cardiac nerve (Boehm and Nussbaum, 1875; Dogiel and Archangelsky, 1906). This twig we have identified, but there appears to be no evidence that it has a vagal effect. The only stimulation of it reported by Boehm and Nussbaum did not alter the heart rate. Notwithstanding this negative evidence we have invariably excluded from action any filament running caudad from the recurrent loop, as well as the filaments passing mediad from the vagus trunk above the heart, by excision of a considerable extent of the common cardiac nerve on the front of the trachea (see fig. 1). After these fine nerve threads and the vagus trunk between the recurrent branch and the heart have been removed, stimulation of the right vagus at the top of the thorax or at various levels in the neck, even as far cephalad as the jugular ganglion, has, in our experience, no effect on the heart. In older literature reference is made to a cardiac connection of the superior laryngeal nerve with the recurrent laryngeal through the anastomosis of Galen (see François-Franck, 1880); we have found no change of heart rate on peripheral excitation of that nerve. Indeed, in the cat, according to our tests, no branches which affect the heart rate are given off from the cervical portion of the right vagus; they arise near the heart.

Our acute experiments and also our observations on numerous cases at the time of surgical operation and at subsequent tests have shown that the cardiac branches leave the *left vagus* trunk near the heart. There is considerable variation, however, in the places of origin. In some cases all the cardiac branches arise close below the recurrent laryngeal branch and no twigs arise from the latter. In other cases important cardiac branches leave the vagus trunk much lower, near the left pulmonary vessels, or pass off from the recurrent laryngeal. Perman (1924) describes a branch which, leaving the left vagus below the hilus of the lung, unites with a similar branch from the right vagus, and from this union a twig passes forward to

the dorsal wall of the auricles. This arrangement, first described by Perman, we have been able to verify in several instances. No cardiac branches arise above the recurrent, as shown in numerous tests by first cutting the recurrent and all other vagal branches below it as far as the junction with the esophagus, then stimulating the left vagal trunk high in the thorax or in the neck, and also the jugular ganglion, and finding no change recorded in the heart rate. Since it is very difficult to cut the fine filaments which may grow out to the heart from the recurrent laryngeal as it turns cephalad under the aortic arch, we adopted the plan of cutting the left recurrent branch itself. Occasionally it runs a relatively long independent course and for that reason may be overlooked. We have definite proof of renewal of vagal connections as early as 68 days after they had been severed (see p. 350), though regeneration probably does not commonly occur so soon.

There has been general agreement that the immediate sympathetic innervation of the heart is derived from the stellate and inferior cervical ganglia. Boehm and Nussbaum (1875), Langley (1892), and Dogiel and Archangelsky (1906) describe only such relations in the cat. In Gaskell's monograph (1920), "The Involuntary Nervous System," the accelerators are described as proceeding from the above mentioned ganglia, and from ganglia on the subclavian ansa. Langley represented all the thoracic nerves from I to V or VI as sending accelerator fibers upward through the sympathetic chain to the stellate, but not as having more direct cardiac connections. And Tigerstedt in his monumental "Physiologie des Kreislaufes" (1921) sums up present knowledge on the basis of Langley's evidence that the relays are in the stellate and inferior cervical stations. According to Boehm and Nussbaum, three sets of fibers are given off from the *right stellate*: two or three prominent strands join the vagus trunk; others pass mediad on the dorsal side of the vagus to join the common cardiac nerve mentioned above; and another strand goes to the esophagus and the hilus of the lungs. Our observations confirm the existence of these sets of fibers. The right inferior cervical ganglion, stimulation of which causes a faster heart beat, gives off branches to the nearby vagus trunk (see fig. 1, and also November 7, 10 and 14, table 1). In one case only out of sixty, Boehm and Nussbaum report finding a connection with a cervical root such as to make the inferior cervical independent. According to Langley the lower cervical spinal nerves of the cat contain no accelerator fibers; those fibers originate in the upper thoracic nerves, chiefly the second and third. The ganglion loses its central connection, therefore, on extirpation of the right stellate, for the preganglionic fibers of the inferior cervical practically always pass through the stellate. In our experience the peripheral connections of the inferior cervical with the heart are destroyed by cutting the right vagus below the recurrent laryngeal

branch; for after that cut the acceleration from exciting the ganglion fails to occur. We have not been able to produce any effect on the heart rate by stimulating the superior cervical sympathetic ganglion, nor have we found by dissection any branches from it to the heart.

A large strand (sometimes two) reaches caudad from the *left stellate*; some twigs from it commonly join the left vagus, while the rest of the strand continues towards the heart. There is also a small fiber, attaching usually at the region of the inferior cervical ganglion, that runs between the in-

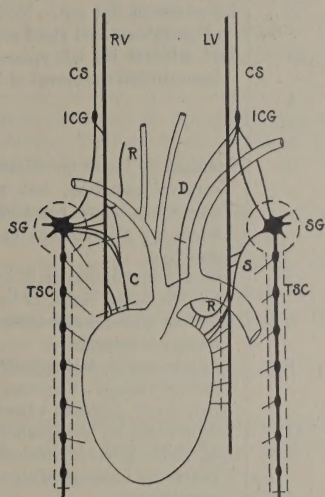


Fig. 1. Diagram of the usual arrangement of cardiac nerves in the cat. *R V*, right vagus; *L V*, left vagus; *C S*, cervical sympathetic; *I C G*, inferior cervical ganglion; *R*, recurrent laryngeal nerve; *D*, depressor nerve; *S G*, stellate ganglion; *C* "common cardiac nerve;" *S*, sympathetic fiber; *T S C*, thoracic sympathetic chain. See text for consideration of fibers reaching mediad from thoracic ganglia below the stellate. The dash lines indicate the parts cut or excised.

nominate and left subclavian arteries, and sends fibers to the ventral side of the aortic arch and onward towards the heart. Sometimes it runs free in the neck. It corresponds to the depressor nerve, as figured by Dogiel and Archangelsky. Although this fiber remained intact in our earlier operations it could not have mediated central or reflex accelerator effects, because removal of the stellate ganglion eliminated the preganglionic connections. When the cervical sympathetic trunk or the superior cervical ganglion was stimulated after the stellate had been removed, no change in

TABLE 1

Records of animals with stellate ganglia removed, vagus nerves cut in the neck, adrenal glands removed, liver nerves severed, and subjected to afferent brachial stimulation after being decorticated and recovering from ether. (Exceptions to these procedures are mentioned under "Remarks")

DATE (1924)	MAXIMUM REFLEX AC- CELERATION OF HEART	REMARKS
	<i>beats per minute</i>	
October 31.	20	Liver nerves not cut. No increase on stimulation of peripheral end right sciatic 30 seconds
November 3.	30	Left adrenal in, left splanchnics cut. Maximum increase after removal of left adrenal, 5 beats
November 4.	5	
November 5.	4	
November 6.	8	
November 7.	0	Increase 28 beats on stimulation of right inferior cervical ganglion, but no increase after right vagus cut below recurrent branch. Increase 4 beats on stimulation of left inferior cervical ganglion
November 10.	2	Increase on stimulation of inferior cervical ganglia; left, 14 beats, right, 22 beats
November 11.	0	Vigorous abdominal massage over liver; increase 2 beats per minute
November 13.	8	Left adrenal in, left splanchnics cut. Ether. No decerebration. Vigorous abdominal massage over liver; increase 4 beats
November 14.	11	Left adrenal in, left splanchnics cut. Stimulation of right inferior cervical ganglion, increase 30 beats; no increase after vagotomy below recurrent branch
December 2, a.m....	20	Left adrenal in, left splanchnics cut 3 weeks previous. Right splanchnic stimulated, no increase
December 2, p.m....	16	Left adrenal in, left splanchnics cut 3 weeks previous
December 4, a.m....	10	Right and left splanchnics cut, liver nerves not cut, both adrenals in
December 4, p.m....	22	Right and left splanchnics cut, liver nerves not cut, both adrenals in. Ether
December 5.	12	Right and left splanchnics cut, liver nerves not cut, both adrenals in
December 6.	24	Light ether, no decerebration, in this and succeeding cases. Splanchnics cut both sides, left upper sympathetic ganglia removed
December 8.	14	Splanchnics cut both sides, liver nerves not cut, adrenals in. At autopsy found 2 large strands from right sympathetic cord to semilunar ganglion
December 9.	6	Splanchnics cut both sides, liver nerves not cut, adrenals in. Upper abdominal sympathetic chain removed both sides

TABLE 1—*Concluded*

DATE (1924)	MAXIMUM REFLEX AC- CELERATION OF HEART	REMARKS
	<i>beats per minute</i>	
December 11.....	24	Splanchnics cut both sides, liver nerves not cut, both adrenals in. After removal of upper abdominal sympathetic chain both sides, reflex increase only 6 beats (168-174); after adrenals tied off, increase, 0
December 18.....	6	Right adrenal removed, left adrenal in, left splanchnics cut, upper left sympathetic chain removed

It should be clearly understood that the operative procedures in these cases left the animals with a satisfactory blood pressure, i.e., above the critical level. The heart rate was recorded on the blood-pressure record.

the heart rate was recorded. Apparently there is no cardiac innervation above the inferior cervical sympathetic ganglion.

The information detailed above naturally led us to conclude that the following operations would assure a heart deprived of all nervous connections with the central nervous system, and that this condition would prevail for possibly two months or more:

1. Removal of both stellate ganglia, thus disconnecting also the inferior cervical ganglia.

2. Excision of several centimeters of the right vagus below the recurrent laryngeal branch and all twigs passing medially from the vagus, as well as a stretch of the common cardiac nerve.

3. Cutting the recurrent and all other branches of the left vagus between the recurrent and a point below the junction of the vagus with the esophagus.

Known factors affecting the denervated heart. Previous studies have furnished evidence that secretions from the adrenal medulla (Cannon and Rapport, 1921; Cannon and Carrasco-Formiguera, 1922), from the liver (Cannon and Uridil, 1921), and from the thyroid gland (Cannon and Smith, 1922) are each capable of accelerating the beat of the denervated heart—the first two within a few seconds after stimulating the nerve supply, the last after a lapse of minutes and then slowly. The nerves effective in producing these responses belong to the sympathetic division. In removing the stellate ganglia the sympathetic distribution to the thyroid gland would be destroyed. If thereafter the nerves to the liver are cut, and one adrenal gland is removed and the nerves to the other are severed, endocrine agencies influencing the heart rate should be eliminated. Then increase of carbon dioxide in the blood, or asphyxia if sufficiently great,

might be expected to make the heart beat more slowly (Patterson, 1915; Cannon, 1919); and rise or fall of temperature would cause the rate to rise or fall correspondingly (Knowlton and Starling, 1912). If in addition to the influences of the organs mentioned above asphyxia and changes of temperature are ruled out, all the known physiological factors which might influence the activity of the denervated heart would be excluded. The heart then should not be much disturbed, certainly not remarkably accelerated, by the activity of the animal.

To our great surprise the expectation just expressed was not realized in animals surviving for some time after the operations. A few examples will reveal the character of the difficulty which confronted us. The testing of these and other cases consisted in counting the heart rate (by stethoscope), or recording it, while the animal was quiet in the lap, and then placing the animal back downward on a comfortable holder, with the fore legs held by straps and the neck enclosed loosely between two vertical posts united above by a cross bar. Usually sooner or later the animal would begin to pull or jerk about ("struggle") and show signs of rage. This exhibition rarely lasted more than a minute but it was likely to be repeated. If the animal did not become spontaneously active, extending the hind legs moderately, or temporarily closing the nares, would commonly induce the reaction.

Cat 233 had its heart denervated in the manner above described on December 31, 1923. On January 28, 1924, the right adrenal was removed and the left splanchnics cut. When tested February 19, the heart rate increased 28 beats; April 19, 114 beats; May 3, 74 beats—107 to 181 per minute. May 10, the liver nerves were severed. June 16, activity increased the rate from 108 to 184—76 beats per minute. June 24, the left adrenal medulla was sucked out; later the same day activity increased the heart rate 114 beats. July 22, in an acute experiment under light ether, the left vagus was found effective on the heart; but, nevertheless, after both it and the right vagus had been cut, afferent brachial stimulation still accelerated the rate reflexly 46 beats per minute.

The heart of cat 244 was denervated February 4, 1924. February 16 the liver nerves were severed. The right adrenal was removed and the left splanchnics cut April 12. April 24, activity increased the rate 140 beats, from 108 to 248; that afternoon the left splanchnics were again inspected and broken to make sure of their exclusion. April 30, struggle increased the rate from 120 to 244—124 beats per minute. May 3, the vagi were tested in the neck, with no effect on the heart rate. June 16, activity increased the rate 80 beats per minute, 144 to 224. June 19, the left adrenal medulla was sucked out. That afternoon the increase was 84 beats per minute, 160 to 244. In an acute experiment under ether, July 11, both vagi were found effective; but after they were cut in the neck, reflex brachial stimulation increased the rate 28 beats per minute. And when the circulation was confined anterior to the diaphragm, brachial stimulation still accelerated the heart 34 beats, from 204 to 238.

The cardiac acceleration, displayed in the foregoing illustrative cases, after all known nervous and humoral factors had been excluded, proved

to be a very disconcerting and mysterious phenomenon. It was clear that unless it could itself be explained and excluded it might be present whenever any of the known factors mentioned above were acting and thus would vitiate any inferences that might be drawn concerning their effects. We endeavored, therefore, to learn the cause of this belated and unknown accelerating agent.

The search for the unknown accelerator. The appearance of a not previously recognized accelerating agent raised the question of the possibility of finding some indication of its presence in acute experiments. In most of our previous acute experiments on medulliadrenal secretion the animals had been under a general anesthetic; on the other hand, when the unknown accelerator was most effective the animal was not under an anesthetic. It seemed possible that the depth of anesthesia would account for the difference. Accordingly, experiments were planned in which the heart would be denervated under ether, the liver also denervated, the adrenal glands removed or inactivated, the hemispheres destroyed or decorticated quickly through the orbits, and then the anesthetic withdrawn. In all, 14 experiments of this general type were performed. The facts which were newly demonstrated or again emphasized by these experiments were as follows:

1. After removal of the adrenal glands reflex stimulation *may* produce marked acceleration of the heart if the nerves of the liver are intact (October 31, table 1). Therefore an accelerator factor may be given off to the blood from that organ. Section of the hepatic nerves, however, excludes that possibility as a reflex or central nervous effect (Cannon and Uridil, 1921).

2. After the adrenal glands have been removed and the hepatic nerves severed, reflex acceleration of the denervated heart is slight and not readily repeated; in 7 such instances (November 3 to 11, table 1) the maximal reflex increase of rate was 8 beats per minute (1 case, November 6), usually it was much less (4 and 5 beats in 3 cases, November 3, 4 and 5; 2 beats or less in 3 cases, November 7, 10 and 11) (cf. Cannon and Rapport, 1921).

3. If only the splanchnic nerves, major and minor, have been cut, the adrenal medulla is not thereby denervated; strands pass to the semilunar ganglia from upper lumbar sympathetic ganglia on each side (November 13 to December 8, table 1; see also December 11). In 10 such instances the reflex increase of heart rate varied between 8 and 24 beats per minute with only the splanchnics cut: when in addition the upper lumbar sympathetic chain was removed, from the diaphragm to the kidneys, the maximal reflex increase was 6 beats per minute (December 9, 11, 18, table 1) (cf. Stewart and Rogoff, 1917). In other words the conditions were like those seen when the liver nerves are severed and the adrenal glands removed.

4. In the cat there is no crossed influence of the splanchnic impulses of one side on the adrenal medulla of the other side. This was shown not only in acute experiments, but also by testing the right major splanchnic nerve weeks after removal of the right adrenal gland and section of the left splanchnic trunk (see December 2, a.m., table 1) (cf. Elliott, 1912; Stewart and Rogoff, 1916).

The foregoing results confirm earlier observations, made during acute experiments, on the efficacy of nerve section as a means of ruling out hepatic and medulliadrenal activity, and leaving only a small acceleration of the denervated heart. The unknown accelerator did not appear in acute experiments. The secondary large acceleration, which develops in surviving animals after all recognized humoral and nervous agencies affecting the heart rate have been eliminated, is left, therefore, unexplained.

It seemed possible that such long periods between stages of denervation as were common in the early experiments, and as are illustrated in the two cases described on page 334, would permit regrowth of nerve connections at the point first denervated (e.g., the heart) at or near the time when the second denervation was performed (e.g., the adrenals). Obviously this chance for error could be eliminated by performing the complete denervation at once or in two operations near together. This was done in a series of cases, as shown in table 2. The basal rates were those occurring when the animal was quiet in the lap; the increases were due to struggle in the manner already described. It is noteworthy that the increases were small shortly after the operation and became larger as time passed (see especially nos. 172, 16 and 28, table 2). In another animal thus operated upon the rate was 153 beats per minute, when quiet, 24 hours after the operation; there was a maximal increase (recorded by string galvanometer) of only 4 beats per minute when the animal became excited and active. Ten days later, however, the increase was 14 beats per minute; the next day thereafter 18 beats, and the day after that 42 beats. As shown by the results in table 2, after elimination of every known humoral and nervous factor such increases of heart rate as 79, 80 and 84 beats per minute would occur after a struggle. When these results were compared with those which followed corresponding operative procedures in the cases of table 1, the situation became very puzzling indeed. Why should the phenomenon occur in surviving animals and not during an acute experiment?

Among possible explanations for the larger increments of rate of the denervated heart in surviving animals was such a change in the isolated muscle in time as to render it sensitive to influences which do not primarily affect it. A rise of blood pressure does not to a noteworthy degree change the rate of the freshly denervated heart (see Cannon and Rapport, 1921). Can it alter the rate of the denervated heart after a lapse of days? Three days after the denervation the blood pressure was raised, in an acute

TABLE 2

Records of increase of heart rate with activity in animals in which stellate ganglia were removed, right vagus cut below the recurrent branch, left vagal cardiac branches severed, right adrenal gland excised, left splanchnic nerves cut, upper left abdominal sympathetic chain removed, and liver nerves severed under aseptic precautions, in a single operation, or in two operations, as indicated. (Exceptions to the foregoing procedures are mentioned under "Remarks")

CAT NO.	DATE OF DENERVATION		HEART RATES			REMARKS
	Heart	Adrenals and liver	Date	Basal	Increase	
172	Oct. 21, '24	Oct. 15, '24	Oct. 22	144	20	Left adrenal medulla sucked out, Oct. 15
			Oct. 23	156	28	
			Oct. 24	141	51	
			Oct. 25	143	79	Died, Oct. 29, infection
2	Dec. 13, '24	Jan. 15, '25	Jan. 20	150	58	Died, Jan. 22
16	Jan. 23, '25	Jan. 23, '25	Jan. 24	134	6	<i>Acute expt.</i> ; ether, reflex stimulation
			Jan. 26	128	60	
			Jan. 26	180	16	
				180	20	
				178	16	
				182	28	
20	Jan. 30, '25	Jan. 30, '25	Feb. 6	204	48	<i>Acute expt.</i> ; ether, vagi cut in neck
				188	40	After left adrenal tied off
26	Feb. 19, '25	Feb. 19, '25	Feb. 25	120	60	Increase under atropine
			Feb. 26	120	60	
			Mar. 2	100	80	
			Mar. 3	164	52	After left vagus cut in neck
			Mar. 4	148	12	<i>Acute expt.</i> ; decortication
				164	0	Stimulation right splanchnic
28	Feb. 21, '25	Feb. 21, '25	Feb. 25	128	20	Increase under atropine
			Mar. 2	124	52	
			Mar. 10	116	84	
			Mar. 11	116	44	
				124	52	
			Mar. 12	120	72	
				132	44	After left adrenal removed

experiment, from 100 to 168 by sudden pressure on the abdomen; the heart rate changed from 178 beats per minute before to 174 during the higher pressure. A similar result was obtained repeatedly in another instance seven days after the denervation. In both cases the animals were decorticated under ether and the tests made after the effect of ether had disappeared. Increased blood pressure did not explain the difficulty.

When an animal struggles muscle metabolites are set free. They have no marked accelerating influence on the rate of the freshly denervated heart (Cannon, Linton and Linton, 1924). They likewise have no accelerating influence when time has passed after the operation. Tests made by stimulating the peripheral end of the cut sciatic in such acute experiments as those just described, 13 and 17 days after the denervation of the heart and adrenals, proved that metabolites do not increase the rate under these circumstances, and therefore that was not the explanation of the mystery.

Again, might not the outlying neurons of the vagus path, still left in place in the heart muscle, assume an autonomous tonic activity when released from control by superior vagus neurons? Consonant with that possibility were paralytic secretion of the submaxillary gland developed after section of the chorda tympani (Bernard, 1864), and the gradually increased tone of the stomach after cutting its vagal supply (Cannon, 1906). Thus the faster rate of the denervated heart might be due to an agent, possibly an internal secretion, inhibiting vagal tone. In our experiments, however, there was no clear indication of the appearance of independent tonic activity of the intracardiac vagal neurons. The rate when the animal is quiet is likely to be somewhat slower as the days pass after the cardiac denervation and the exclusion of hepatic and adrenal influences, but an amount of atropine which fixates the iris neither causes a faster rate nor prevents the acceleration due to struggle (see nos. 26 and 28, table 2). Since atropine blocks vagal impulses, the theory of an inhibited vagus tone had to be surrendered.

The agencies which remained could be classified into endocrine and nervous. The acceleration was too prompt to be due to thyroid influence. Furthermore, it occurred in an animal with thyroids as well as parathyroids removed, in addition to excision of one adrenal (the right) and denervation of the other and also of the liver (see no. 28, table 2). At one time we thought it might be due to an influence from the pituitary, because brief closure of the cerebral vessels was followed by a faster heart rate in an animal deprived of all known accelerator factors. This possibility was made improbable, however, by an experiment in which the cord was transected between the first and second dorsal vertebrae in an animal whose heart, deprived of all known accelerators, had nevertheless been accelerating as much as 76 beats per minute; though the anterior part of

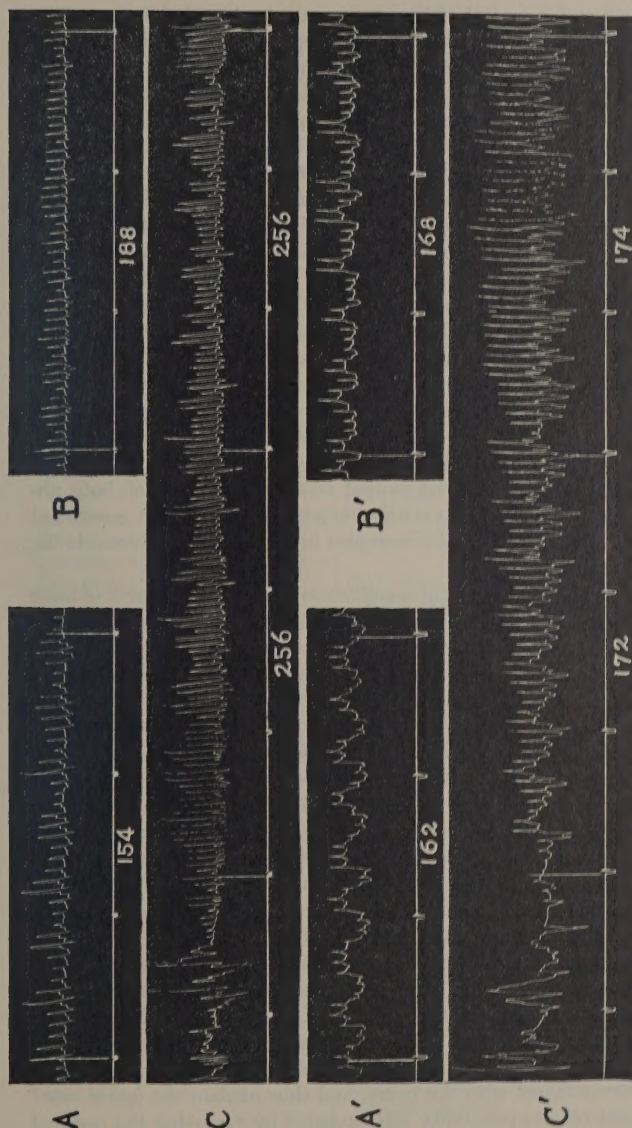


Fig. 2. Records of the heart beat of cat 95 (see table 3). November 2 (a.m.), 1925, after removal of all previously known accelerating agents, the heart rate, when the animal was quiet on the observer's lap, A, was 154 beats per minute; quiet on the animal holder, B, 188 beats; after struggle (indicated by irregular record) C, 256—an increase of 102 beats. January 6, 1926, after removal of upper thoracic sympathetic chains on both sides (November 2, p.m. and December 8), the heart rate, quiet on lap, A', was 162; quiet on holder, B', 168; and after vigorous struggle C', 174—an increase of 12 beats. Time marked in 5-second intervals.

the body, still under control of the brain, was capable of showing excitement and struggle, the heart rate, which dropped from about 120 to 108 beats, could not be made faster by more than 8 beats per minute. It seemed probable, therefore, that the disturbing factor was in the thorax or abdomen. Of the abdominal endocrine organs the gonads were hardly to be suspected of effects on the heart rate. The pancreas had already been shown to be without influence (Cannon and Uridil, 1921). There remained accessory adrenal tissue. This seemed a quite possible cause of the faster beat. The chromophil threads and ribbons associated with the abdominal sympathetic structures contain active adrenin (Vincent, 1910). They are supplied with nerve filaments. It is well known that the iris, deprived of its sympathetic innervation, becomes gradually more sensitive to adrenin secreted into the blood stream; might not the denervated heart likewise become sensitive as time passed? And might not the accessory adrenal tissue gradually hypertrophy because of a deficient supply of adrenin from the main sources? Thus the gradually increasing responsiveness of the heart to excitement and struggle could be reasonably explained. This plausible theory was made untenable when, after inactivation or destruction of the accessory chromophil tissue by removal of both abdominal sympathetic chains in a series of experiments, the heart continued to accelerate just as before. As examples in this series, see records 92, 95 and 110 in table 3.

All possible abdominal causes of acceleration that we could think of have now been reported as having been tested and excluded, except massage of the liver or the remaining adrenal medulla. Actual massage of the liver, more vigorous than would result from contraction of abdominal muscles, failed to reproduce the marked rise of heart rate seen when the animal moved vigorously (cf. November 11 and 13, table 1). And the adrenal factor was excluded, after the liver had been denervated, the right adrenal had been removed and the left adrenal medulla had been sucked out through a cut along one edge of the gland, by observing that the heart could still be made to beat faster (see cases, p. 334 and also no. 172, table 2). The operation was performed with aseptic precautions and allowed repeated tests to be made after the animals had fully recovered their vigor. Thus the evidence pointed to the accelerating agent being below the neck, but not in the abdomen (cf. also cat 244, p. 334).

In the thorax no humoral accelerating factor could be conceived. The cause must be nervous. The stellate ganglia had been removed; they could not mediate the accelerator impulses. Langley (1897) has pointed out a fairly quick renewal of sympathetic connections. Might not pre-ganglionic fibers grow out soon to the inferior cervical ganglia, and might not their axons connect with the heart, and thus explain the faster rate? This somewhat remote possibility was excluded by removing the cervical

TABLE 3

Records of animals in which all hitherto known humoral and nervous accelerators of the heart were removed, and part or all of the abdominal sympathetic chain (cases 92, 95, 97 and 110), without eliminating the faster beat of the denervated heart. Only small increases of heart rate occurred in these cases, and in 4 and 23, when more of the thoracic sympathetic chain was removed. In every instance a section of the common cardiac nerve and of the right vagus trunk below the recurrent laryngeal branch was excised, and the cardiac branches of the left vagus were cut

NUMBER AND DATE	HEART RATE		OPERATIONS AND REMARKS
	Basal	Increase	
(92) 1925			
September 4. . . .			Right thoracic sympathetic chain removed from ribs 1 to 3; left, 1 to 4
September 18. . . .			Left splanchnic and liver nerves cut, right adrenal and upper 3 left abdominal sympathetic ganglia removed
September 22. . . .	117	36	
October 6.			Abdominal sympathetic chains removed
October 15.	112	36	
October 21.			Right splanchnics cut
October 24.	112	24	Animal in poor condition
November 25. . . .	156	34	Animal in better health
December 2.	144	38	
December 8.			Right thoracic sympathetic chain removed from ribs 3 to 9; left, 4 to 9
December 9.	100	4	
December 12. . . .	110	6	
December 14. . . .	100	14	
December 18. . . .	130	10	
December 21. . . .	128	12	
December 31. . . .	158	6	
1926			
January 6.	152	8	
January 19.	142	24	
January 29.	154	26	
(95) 1925			
September 4. . . .			Right thoracic sympathetic chain removed from ribs 1 to 3; left, 1 to 4
September 15. . . .			Left splanchnic and liver nerves cut, right adrenal and upper 3 left abdominal sympathetic ganglia removed
September 16. . . .	152	32	
September 29. . . .	152	72	
October 5.	148	73	
October 13.	146	84	
October 14.			Right splanchnics cut, abdominal sympathetic chains removed
October 19.	156	84	

TABLE 3—Continued

NUMBER AND DATE	HEART RATE		OPERATIONS AND REMARKS
	Basal	Increase	
November 2....	154	102	Remnant of right thoracic sympathetic chain completely removed. (Operation after observation)
November 7....	176	58	
November 18...	172	34	
December 2....	176	36	
December 8....			Left thoracic sympathetic chain removed from ribs 4 to 9
December 9....	152	9	
December 14....	182	12	
December 18....	186	12	
December 21....	172	4	
December 31....	180	8	
1926			
January 6.....	162	12	
January 18.....	144	16	
January 29.....	172	22	
(110)			
1925			
September 19...			Right thoracic sympathetic chain removed from ribs 1 to 8, left stellate out
October 14.....			Left splanchnic and liver nerves cut, right adrenal and upper 3 left abdominal sympathetic ganglia removed
October 19.....	137	83	
November 2....	164	72	
November 3....			Right splanchnics cut, abdominal sympathetic chains removed
November 18...	183	49	
December 2....	172	65	
December 3....			Left thoracic sympathetic chain removed from ribs 2 to 9
December 4....	116	8	
December 6....	152	8	
December 9....	144	4	
December 14....	124	14	
December 21....	136	8	
December 31....	146	30	
1926			
January 6.....	136	18	
(97)			
1925			
July 31.....			Right thoracic sympathetic chain removed from ribs 1 to 4; left, 1 to 8
August 10.....			Left splanchnic and liver nerves cut, right adrenal and upper 4 left abdominal sympathetic ganglia removed
August 12.....	120	8	

TABLE 3—*Concluded*

NUMBER AND DATE	HEART RATE		OPERATIONS AND REMARKS
	Basal	Increase	
August 13.....	99	24	Lower right thoracic sympathetic chain removed. (Operation after observation)
August 15.....	104	30	
August 20.....	132	24	
August 21.....	88	8	
September 1....	92	12	Left splanchnic and liver nerves cut, right adrenal and both abdominal sympathetic chains removed
September 10... (4) 1925	100	10	
September 30...			
November 28...			
November 30...	140	4	Right and left thoracic sympathetic chains removed from ribs 1 to 8
December 2....	152	12	
December 5....	144	4	
December 9....	142	8	
December 12....	116	10	
December 18....	120	12	
December 21....	106	10	
December 31.... 1926	120	8	
January 6.....	126	14	
January 29..... (23) 1926	152	32	
January 21.....			
February 11....			
February 12....	76	14	Right and left thoracic sympathetic chains removed from ribs 1 to 8; liver nerves cut
February 15....	78	6	
February 17....	88	8	
February 19....	86	8	
February 23....	104	8	Left splanchnic nerves cut, right adrenal and upper 4 left abdominal sympathetic ganglia removed
February 25....	78	10	
March 1.....	114	6	

sympathetic chain on both sides in three animals from which all other known accelerators had already been excluded. Struggle hastened the heart rate quite as much as before.

There remained only the possibility of accessory accelerator fibers from

thoracic sympathetic ganglia below the stellates. As early as November 3, 1924, the promptness of acceleration of the heart, though freed from recognized accelerator influences, suggested that a nervous agent was operating. And on February 6, 1925, when under similar conditions an increase of 40 beats per minute was recorded (see no. 20, table 2) and at autopsy a nerve strand was found passing from a left thoracic ganglion to the arch of the aorta, the possibility seemed confirmed. Accordingly, thereafter the thoracic sympathetic chain was removed down to the third intercostal space on the right side and to the fourth or fifth on the left side. In spite of this addition to the standard operation the acceleration persisted when theoretically it should not. One of these cases manifested an increase of 106 beats per minute when the animal passed from a quiet to an excited and active state, though all splanchnic nerves had been severed, the celiac ganglion excised in addition, the right adrenal gland removed and the left adrenal medulla sucked away! The left thoracic sympathetic chain had been taken out from the stellate ganglion down to the sixth rib inclusive. An extraordinary change occurred in this animal when the right thoracic sympathetic chain was further removed from the third to the ninth intercostal space. The quiet heart rate promptly fell from about 150 beats per minute to about 100, and the increase of rate on exciting the animal fell from 106 to 4. Further decisive evidence was obtained in another case in which the right thoracic chain had been taken out down to the eighth rib and the humoral accelerators eliminated (cat 110; table 3 and figure 3). On the left side the stellate only had been removed. Excitement increased the heart rate as much as 83 beats per minute. Now the left thoracic chain was removed from the second to the ninth rib. The quiet rate dropped about thirty beats and the increase resulting from excitement dropped from the region of 65 to 8 and 14. In still another instance (cat 92, table 3 and fig. 3), in which the quiet rate was about 140 and the increment when the animal struggled was about 40 beats per minute, the right thoracic chain was removed from the third to the ninth rib and the left from the fourth to the ninth; the basal cardiac rate fell to 100 to 110, and the increment fell to 4 to 14 beats per minute. Cat 95, table 3 and figure 3, illustrates further the effect of eliminating the upper thoracic ganglia below the stellates (see also fig. 2). Initial removal of the thoracic ganglia, down to the eighth rib, on both sides in one operation (see cats 4 and 23, table 3 and fig. 3) confirmed these results. With the humoral factors excluded the maximal increase in case 4 during the first four weeks was 12 beats per minute—a minor effect as compared with 70 or 100 beats. Other similar cases have yielded still further support for the conclusion that the mysterious increment of rate was due to accelerator fibers reaching the heart from the sympathetic chain below the stellate ganglia, and even

below the third or fourth thoracic ganglia.⁴ More evidence will be reported in later papers.

Perman (1924), in a recent exhaustive article on cardiac innervation, has described fibers passing to the heart in the calf from ganglia III to VI of the left thoracic sympathetic chain, and from ganglia III and IV on the right side, besides the fibers from the stellate ganglia. Similar fibers are known to proceed in man from the upper five or six thoracic ganglia to the cardiac plexus. Although Perman, and Reighard and Jennings (1901) do not record any fibers passing mediad from the sympathetic chain in the cat, it is interesting to note that Mivart (1881) described fine branches given off by the first five or six thoracic ganglia of the cat and distributed "mostly to the aorta and the adjacent parts." The indication that accelerator influences reach the heart from thoracic ganglia below the stellate led us to dissect carefully and with the aid of a dissecting microscope the upper part of the sympathetic chain in more than thirty animals. In nearly all of them fine fibers could be seen passing toward the mid-line from most of the first three or four ganglia below the stellate on either side and occasionally from the fifth and sixth. As a rule it was difficult to trace these fibers to the heart, but in one instance a larger strand than usual was followed to the aortic arch, and in another instance a well defined strand was found connecting the ganglion below the right stellate with the root of a pulmonary vessel.⁵ It seems fairly certain, therefore, that upper

⁴ This conclusion with the evidence supporting it was presented at a meeting of the Harvard Medical Society, January 12, 1926 (see *Boston Med. and Surg. Journ.*, 1926, exciv, 187). The account detailed above gives no indication of the very great difficulties encountered in the course of the investigation. The technical troubles incidental to operating successfully on the chest were themselves serious. Furthermore, after animals had recovered well from operation, they often were afflicted with an epizootic of "snuffles" which made them ill for weeks or caused their death. Thus much valuable time and effort were lost. The minor acceleration immediately following the denervation of the heart, and its gradual development thereafter were most baffling. Naturally the cause could not be discovered by acute experiments. In our bewilderment we had to consider every possible reason for the faster rate and to test every possible agency. The search took, perhaps, much longer than it should. It involved the coöperation of many workers, as indicated by the names at the beginning of this article. Although the "authors" brought the work to a conclusion, the "cooperators," especially J. R. Linton and R. R. Linton, made important contributions to the final outcome.

It is of interest to note that under certain conditions after removal of all the accelerating agencies, struggle caused the heart to beat more slowly. This phenomenon will be considered in a later communication.

⁵ Confirmation of these observations was reported in a paper by Dresbach and Waddell (*Journ. Pharm. and Exper. Therap.*, 1926, xxvii, 9) which appeared after our paper was prepared for publication. They found connections between the cardiac plexus and the thoracic sympathetic chain as far down as the fifth ganglion. They

thoracic ganglia other than the stellate send accelerator fibers to the heart, though the fibers are often too small to be readily traced to their destination.⁶ In any case the physiological evidence was clear, and the need for removing the upper sympathetic chain in the thorax was evident, if a completely denervated heart was desired. We are now in a position to describe the procedure of denervation.

Procedure of cardiac denervation. Vigorous young male or non-pregnant female cats are selected. After the animal has been deeply etherized a sterilized rubber catheter, 3 mm. in diameter, cut slantingly at the end, is passed through a hole in a wooden stick and introduced into the trachea. The glottis is exposed by separating the jaws with cords slipped between the teeth, and by pulling the tongue forward. A good illumination is necessary for quick action before the deep anesthesia lifts and laryngeal reflexes occur. The stick is placed back of the canine teeth and held there by a thong or strong tape tied back of it and around the jaws. The catheter, reaching to a point a short distance above the bifurcation of the trachea, is fixed in place by a thread fastened tightly around it and now tied to the stick. Thereafter the animal is given artificial respiration by means of an interrupted air blast. The air passes through an ether bottle so arranged that the amount of ether which is added can be nicely regulated. After leaving the ether bottle the air-ether mixture is forced, in cold weather, through a coil of copper tubing which is set in warm water. Thus the air is delivered in the trachea at a temperature approximately normal for natural breathing.

did not present any evidence as to whether these fibers were afferent or efferent. In our experience the heart will not be with certainty completely deprived of sympathetic connections, as they state, if the thoracic chain is removed only "as low down as the fourth or fifth ganglion."

⁶ Several questions remain to be answered. It is not clear why the heart should accelerate only slightly in acute experiments, and during the first day or two after surgical removal of stellate and vagal influences and exclusion of humoral accelerator factors, and then should gradually respond more and more, as the days pass, to conditions which excite sympathetic activity. It seems possible that removal of the stellate ganglia damages the thoracic chain below the stellates so that it does not function perfectly until it has recovered. Or the heart may be affected by ether and trauma so that it is not so sensitive as it is otherwise. Or possibly the thoracic fibers below the stellate take on a gradually increasing compensatory service after the principal stellate control has been abolished. For the present, however, this question is left unanswered.

Another interesting question is the variation in the basal rate of the denervated heart in different animals and in the same animal at different times (see table 3). There is no indication that the slower rate is associated with persistent vagal effects. This question is set aside for future answer.

A third problem is that of the slight increase in heart rate (4 to 14 beats per minute) after known endocrine factors have been suppressed and thorough cardiac denervation has been performed. Possibly warmer blood brought to the heart would explain the faster rate. We do not now offer, however, any evidence on the point.

The hair is closely clipped from the right and left chest over the fifth intercostal space. These regions are first thoroughly scrubbed with soap and water, next flushed with 95 per cent alcohol, and then with ether; when nearly dry they are wet with a 7 per cent tincture of iodine. Except in these two regions the animal is covered with sterile towels, which in turn are covered with a sterile sheet having an aperture placed over the field of operation. The skin is now incised over the fifth interspace on the right side, and sterile cloths are clipped to the edges. The superficial

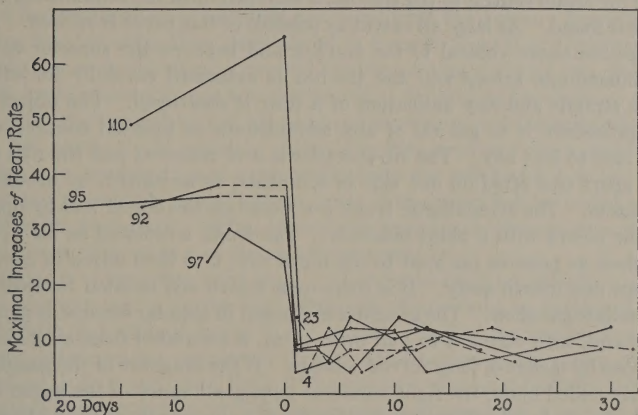


Fig. 3. Graphic representation of the cases reported in table 3. After all hitherto known accelerating agents had been removed the increases of rate of the denervated heart were sharply and markedly reduced in cases 92, 95, 97 and 110 by removal of the remnants of the thoracic sympathetic chain down to the eighth or ninth rib. In cases 4 and 23 (represented by dash-lines) removal of the humoral and nervous accelerating factors, including initial removal of both thoracic sympathetic chains from the first to the eighth ribs, resulted from the outset in only slight increments of the heart rate.

muscles are cut across parallel to the interspace, and the intercostal muscles are broken through with a blunt hemostat while the artificial respiration is temporarily stopped. A sterile silk cloth is introduced to prevent the lung from covering the area of operation at each inspiratory blast, and to protect it from being injured. The fifth and sixth ribs are pushed apart by means of a rib-spreader, and as the spreader widens the gap the intercostal muscles are cut until the opening, which should be extended well toward the back, is about 3 cm. wide. Under protection of a silk cloth the lung anterior to the opening is pressed caudad and is held posterior to the opening and towards the ventral side by means of a flexible retractor inserted between

the arms of the rib-spreader. Thus the fore part and the dorsal aspect of the thoracic cavity are exposed to view.

By means of a blunt dissector, the vagus nerve is uncovered as it courses alongside the trachea. It is grasped in forceps, severed close to the azygos vein, then traced forward to the junction of the accelerator strands from the stellate, and severed again. The part thus isolated is removed. Thus also is excised any branch to the heart that may arise from that portion of the trunk, and the possibility of regrowth of the trunk is minimized. Next the tissue ventral to the trachea is searched until the common cardiac nerve is found. As large an extent as possible of this nerve is excised. All connective tissue ventral to the trachea and between the superior cava, the innominate artery and the trachea is examined carefully for other nerve strands and any indication of a fiber is destroyed. The object of this procedure is to get rid of any adventitious or unusual connections. It is rare to find any. The rib-spreader is now removed and the ribs are held apart and lifted on one side or the other, as necessary, by means of retractors. The sympathetic trunk is exposed on the dorsal wall by opening the pleura with a blunt dissector. The trunk is isolated as far down the chest as possible (at least to the ninth rib); it is then seized in curved forceps and drawn away. It is thereupon traced and isolated forward to the stellate ganglion. The ganglion is grasped in angular forceps, is gently lifted and, with care not to pull upon them, is separated from all its connections by means of long curved scissors. If the branches of the ganglion are not pulled upon, the right recurrent laryngeal branch of the vagus will not be injured and will remain to care for the movements of the right side of the larynx. Transection of the right vagus and the common cardiac nerve and removal of the right thoracic trunk I to VIII have now denervated the heart on the right side. After the silk has been taken out and a slightly increased air pressure has inflated the collapsed lung, the chest is closed, first by a loop of linen thread drawing into their normal positions the fifth and sixth ribs, and then by sewing the different muscle layers together with black silk. The wound is closed by a continuous silk suture, carried in a fine needle, which is pressed through the skin as close as possible to the edge of the cut. A dry dressing of sterile cotton or gauze is temporarily fastened over the wound with strips of adhesive tape.

On the left side a similar opening is made in the fifth intercostal space. The upper lobe of the left lung is gently pressed caudad by means of a silk cloth and it and the other lobe of this region, protected by another silk cloth, are held posterior to the opening and toward the *ventral* side of the chest by means of a curved retractor. The left vagus nerve is uncovered at the point where it joins the esophagus. From that point forward all its branches are cut, up to and including the recurrent laryngeal. The small fiber (depressor) mentioned on page 331 is next severed. The vagus is

then tested electrically in order to prove that all cardiac branches have been disconnected. When that is assured the rib-spreader is removed, the chest wall is lifted by means of retractors, and the left sympathetic chain is removed from the stellate ganglion, inclusive, to the ninth rib, as on the right side. After the silk cloths have been removed and the lung well inflated (it may be necessary to unfold the anterior lobe gently with a blunt dissector), the chest is closed as on the right side. The intratracheal tube should not be withdrawn until the animal is breathing naturally.

The after-care of the animals was directed towards keeping them warm and isolated until they had fully recovered from the ordeal. Immediately after the operation they were placed in a small cage; if the weather was cold they were placed on a covered electric heating pad and also provided with heat reflected from a hot electric coil. By varying the distance of the reflector the temperature of the cage could be quite satisfactorily adjusted to the needs of the animal. After complete recovery from anesthesia water was presented; on the second day milk was offered; and on the third day and thereafter canned salmon, raw liver or cod fish or boiled meat, with milk, were given. When the animals were again in vigorous condition they were allowed to run about a large room.

Methods of recording heart beats. In order to have an objective record of the heart rate a device was necessary which could be employed conveniently and without injury to the animal. At first use was made of the string galvanometer, connected through brine-soaked gauze with a shaven spot on a fore and hind limb. The records were satisfactory, but the method had the defects of being expensive and requiring a trained technician to operate the galvanometer. The method finally adopted was simple, inexpensive and capable of being operated by the observer himself without help.

The apparatus consisted of receiving and recording tambours connected by about 5 feet of small rubber tubing. The receiving tambour was a thistle tube, about 4 cm. in diameter, the mouth of which was covered with a sheet of thin rubber (dental dam or a piece of rubber glove will serve). The recording tambour was provided with a compound lever and was highly sensitive. One or two small pin holes burnt through the rubber of the recording tambour allowed adjustment of the receiving tambour to different surfaces without impairing the sensitiveness of the system to quick, slight pressure changes.

In applying the apparatus the animal is first placed in the situation selected, quiet in the lap or fastened to a holder, and then, after the region of maximal cardiac impulse against the chest wall has been found by palpation, the receiving tambour is held firmly over it. Thereupon with each impulse the writing lever quickly rises and falls. By registering this motion, and the time, on a revolving drum a record of the heart rate is

obtained (see fig. 2). The records vary at different times because the position of the animal shifts and the receiving tambour cannot always be applied in the same place.

The recording apparatus worked best on the flexible chest walls of young cats, for after the ribs have become stiffened with age the cardiac impulse is often not sufficiently prominent to permit good records to be taken. Both vigor to stand the operation and ease of securing cardiac records, therefore, led to the selection of young animals.

Duration of the denervation. Examination of table 3 reveals that in cases 92, 95, 110 and 4, after a period of relatively slight cardiac accelerations, the heart began to respond by larger increments. After 33 days of no greater increase than 12 beats per minute, no. 4 showed an increase of 14 beats on the thirty-ninth day; it rose to 32 on the sixty-second day and to 60 on the eighty-fourth. In nos. 92 and 95 the greater increments began about 40 days after *completing* the denervation. In these cases, as well as in 110, the operation had been performed in several stages (see table 3). A terminal acute experiment on no. 4 proved that the cardiac denervation, done 89 days previously, was still perfect (neither vagal nor thoracic sympathetic stimulation evoked any change); the right splanchnic, however, caused some acceleration, possibly by renewed hepatic connections, though proof was not secured. In a terminal acute experiment on no. 95, stimulation of the right vagus trunk, cleared of remnants of the cervical sympathetic strand, and at a distance of 4 cm. from any other tissue, caused a momentary decrease of heart rate of 32 beats per minute (from 128 to 96) and thereupon, while the stimulation was continuing, an increase of 40 beats per minute (from 96 to 168). After removal of the inferior cervical ganglion the same stimulation decreased the rate 18 beats per minute, and later increased it only 4 beats. The right thoracic sympathetic chain had been removed more than three months previously; the right vagus had been cut below the recurrent nearly six months. It appears probable that the cut vagus fibers had made new connections with both the cardiac vagus neurons and the inferior cervical ganglion. In another instance we found evidence that after four months the inferior cervical ganglia may reconnect with both the heart and with the central nervous system. It is clear that as time passes after denervation very complicated connections may be established between the heart and the central nervous system.

The foregoing cases belonged to a stage previous to the final development of our technic and should be regarded, therefore, as not showing the probable persistence of the denervation. The same comment may be made regarding two cases in which vagal slowing of the heart was demonstrated 68 days after the vagi were deprived of cardiac influence. Nevertheless, because now it appears that regeneration of cut cardiac nerves

may occur within two months or less and thereby may greatly complicate the responses of the heart, the time of observation must be carefully limited. For the present, therefore, it seems prudent to use the heart as an indicator within 30 days after the denervation, and then, by completely eliminating the humoral factors and finding no marked acceleration thereafter, to prove in each case that the heart has in fact been denervated and has remained so.

SUMMARY

The value of the denervated heart in a surviving animal is pointed out. It is a representative organ, existing in the fluid matrix of the body, performing its normal functions, with activity capable of being easily recorded, and influenced only by changes in the blood which perfuses it.

The recognized innervation of the heart of the cat is described (see fig. 1).

When the hitherto recognized nerves reaching to the cat's heart are severed and all known humoral agents (adrenal glands, liver) are excluded from action, the heart nevertheless becomes capable of markedly accelerating if the animal changes from a resting to an excited, active state.

Details are given of the search for the unknown accelerating agent (see tables 1, 2 and 3 and figs. 2 and 3). After elimination of changes in the heart muscle following denervation, and exclusion of one humoral agent after another, as disturbing factors, evidence is presented showing that accessory accelerator fibers from upper thoracic ganglia below the stellates mediate the faster beat. When these are removed, in addition to the removal of the previously recognized agencies causing the heart to beat faster, the heart rate remains steady within approximately 12 beats in spite of vigorous activity (see table 3 and fig. 3).

The details of the procedure for completely denervating the cat's heart are described, as well as the after-care of the animals.

A method for mechanically recording the heart rate of the cat is described.

BIBLIOGRAPHY

- BENEDICT, F. G. 1915. A study of prolonged fasting. Carnegie Inst. of Washington, Publ. no. 203, 118.
- BENEDICT, F. G., W. R. MILES, P. ROTH AND H. M. SMITH. 1919. Human vitality and efficiency under prolonged restricted diet. *Ibid.*, Publ. no. 280, chart 124.
- BERNARD, C. 1864. *Journ. de l'Anat. et de la Physiol. Norm. et Pathol.*, i, 507.
- BOEHM, R. AND H. NUSSBAUM. 1875. *Arch. f. Exper. Path. u. Pharm.*, iv, 255.
- CANNON, W. B. 1906. *This Journal*, xvii, 436.
1919. *This Journal*, i, 399.
- CANNON, W. B. AND D. RAPPORT. 1921. *This Journal*, lviii, 307.
- CANNON, W. B. AND J. E. URIDIL. 1921. *This Journal*, lviii, 354.
- CANNON, W. B. AND P. E. SMITH. 1922. *This Journal*, lx, 476.
- CANNON, W. B. AND R. CARRASCO-FORMIGUERA. 1922. *This Journal*, lxi, 215.

- CANNON, W. B., J. R. LINTON AND R. R. LINTON. 1924. *This Journal*, lxxi, 153.
- DOGIEL, J. AND K. ARCHANGELSKY. 1906. *Pflüger's Arch.*, cxiii, 45.
- ELLIOTT, T. R. 1912. *Journ. Physiol.*, xlv, 401.
- EVANS, C. L. 1912. *Journ. Physiol.*, xlv, 213.
- FRANÇOIS-FRANCK, C. A. 1880. *Travaux du Laboratoire de Marey*, iv, 75.
- FRIEDENTHAL, H. 1902. *Arch. f. Physiol.*, 135.
- GASKELL, W. H. 1920. The involuntary nervous system, 34.
- GUTHRIE, C. C. AND F. H. PIKE. 1907. *This Journal*, xviii, 28.
- HARRIS, J. A. AND F. G. BENEDICT. 1919. A biometric study of basal metabolism in man. *Carnegie Inst. of Washington*, Publ. no. 279, table 22.
- KNOWLTON, F. P. AND E. H. STARLING. 1912. *Journ. Physiol.*, xlv, 206.
- LANGLEY, J. N. 1892. *Phil. Trans. Roy. Soc. London, Series (B)*, clxxxiii, 107.
1897. *Journ. Physiol.*, xxii, 215.
- MINOT, G. R. AND J. H. MEANS. 1924. *Arch. Int. Med.*, xxxiii, 576.
- MIVART, ST. G. 1892. *The cat*. New York.
- PATTERSON, S. W. 1914-1915. *Proc. Roy. Soc. London, Series (B)*, lxxxviii, 380.
- PERMAN, E. 1924. *Zeitschr. f. Anat. u. Entwicklungsges.*, lxxi, 382.
- REIGHARD, J. AND H. S. JENNINGS. 1901. *Anatomy of the cat*. New York.
- ROGOFF, J. M. 1924. *This Journal*, lxvii, 551.
- SMITH, H. M. 1922. Gaseous exchange and physiological requirements for level and grade walking. *Carnegie Inst. of Washington*, Publ. no. 309, 170.
- STEWART, G. N. AND J. M. ROGOFF. 1916. *Journ. Pharm. Exper. Therap.*, viii, 504.
1917. *Journ. Exper. Med.*, xxvi, 613.
- STURGIS, C. C. AND E. H. TOMPKINS. 1920. *Arch. Int. Med.*, xxvi, 467.
- TIGERSTEDT, R. 1921. *Die Physiologie des Kreislaufes*, ii, 388.
- VIERORDT, H. 1906. *Anatomische, physiologische und physikalische Daten und Tabellen*. 3rd. ed., Jena, 44.
- VINCENT, S. 1910. *Proc. Roy. Soc. London, Series (B)*, lxxxii, 502.

